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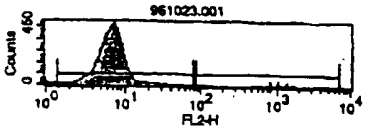
(54) Title: USE OF RAR ANTAGONISTS AS MODULATORS OF HORMONE MEDIATED PROCESSES



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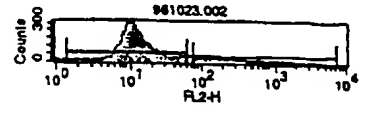

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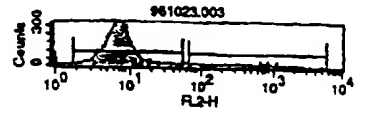

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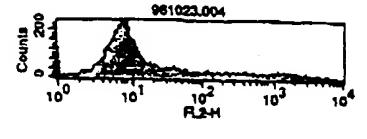

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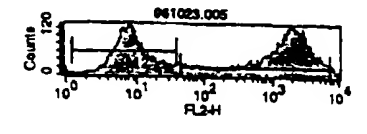

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M2	10.76	8.87	68.26	8.35

(57) Abstract: In accordance with the present invention, it has been discovered that retinoic acid receptor (RAR) antagonists are capable of modulating processes mediated by other members of the steroid/thyroid hormone receptor superfamily, including permissive receptors such as PPARs (e.g., PPARα, PPARδ and PPARγ). Indeed, it has been discovered that RAR antagonists, in combination with agonists for members of the steroid/thyroid hormone receptor superfamily, are capable of inducing and/or enhancing processes mediated by such members.

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Use of RAR Antagonists as Modulators of Hormone Mediated Processes

FIELD OF THE INVENTION

The present invention relates to methods for the modulation of nuclear receptor mediated processes. In a particular aspect, the present invention relates to methods for modulating the activity of members of the steroid/thyroid hormone receptor superfamily by relieving the inhibition of hormone mediated processes caused by retinoic acid receptor, or agonists thereof. In another aspect, the present invention relates to methods for inducing hormone mediated processes.

BACKGROUND OF THE INVENTION

A central problem in eukaryotic molecular biology continues to be the elucidation of molecules and mechanisms that mediate specific gene regulation. As part of the scientific attack on this problem, a great deal of work has been done in efforts to identify ligands (i.e., exogenous inducers) which are capable of mediating specific gene regulation. Additional work has been done in efforts to identify other molecules involved in specific gene regulation.

Although much remains to be learned about the specifics of gene regulation, it is known that ligands modulate gene transcription by acting in concert with intracellular components, including intracellular receptors and discrete DNA sequences known as hormone response elements (HREs). The identification of compounds which directly or indirectly interact with intracellular receptors, and thereby affect transcription of hormone-responsive genes, would be of significant value, e.g., for therapeutic applications.

The actions of steroids, retinoids and thyroid hormones are mediated by intracellular nuclear receptors whose coordinate activity defines the physiological

response (Mangelsdorf and Evans, *Cell* **83**:841-850 (1995)). These receptors are all structurally related and constitute a superfamily of nuclear regulatory proteins that modulate gene expression in a ligand-dependent fashion. Previous studies have demonstrated that the 9-cis retinoic acid receptor (RXR) serves as a common heterodimeric partner for thyroid hormone receptor (TR), retinoic acid receptor (RAR), vitamin D receptor (VDR), prostanoids (PPAR), as well as numerous orphan receptors (Kliewer et al. (1992) *Nature* **355**:446-449).

Nuclear hormone receptor heterodimers can be classified into two distinct groups based upon their transcriptional responses to synthetic RXR ligands. So called "permissive" heterodimers such as PPAR:RXR, respond to either RXR and/or PPAR ligands and the two together have, at least, an additive effect (see, e.g., Mukherjee et al., *Nature* **386**:407-10 (1997)). In contrast, so called "non-permissive" heterodimers, such as RAR:RXR, do not respond to RXR ligands unless ligands for RAR are already present, in which case they yield an additive or synergistic response (Apfel et al., *J Biol Chem.* **270(51)**:30765-72.(1995); Chen et al. *PNAS* **93**:7567-7571 (1996)). Other non-permissive heterodimers include TR:RXR and VDR:RXR heterodimers, which also do not appear to be activated by RXR ligands. Indeed, the RXR ligand, LG100268, appears to partially antagonize the action of thyroid hormone.

This difference between permissive and non-permissive heterodimers is likely to be important for regulating the activity of naturally occurring RXR ligands (Heyman et al., *Cell* (1992) **68**:397-406; Mascrez et al., *Development* (1998) **125(23)**:4691-707; Solomin et al., *Nature* (1998) **395(6700)**:398-402; Fujita and Mitsuhashi, *Biochem Biophys Res Commun* (1999) **255(3)**:625-30)] as well as being crucial to understanding the behavior of synthetic compounds currently under development as both anti-cancer and anti-diabetic agents (see, e.g., Anzano et al., *Cancer Research* (1994) **54**:4614-4617, Gottardis et al., *Cancer Research* (1996) **56**:5566-5570, Mukherjee et al., *supra*). It has been also suggested that RXR can function as a homodimer (Mangelsdorf et al., *Cell* **66(3)**:555-61 (1991)). By

competing for dimerization with RXR on response elements, the relative abundance of RAR and PPAR determines whether the RXR signaling pathway will be functional.

PPARα is a permissive member of the nuclear receptor superfamily, which includes receptors for the steroid, thyroid and retinoid hormones (see Mangelsdorf & Evans in *Cell* **83**:841-50 (1995)). Two other PPARα-related genes (PPARγ and PPARδ) have been identified in mammals. PPARγ is highly enriched in adipocytes, while the δ isoform is ubiquitously expressed (see Schoonjans et al., in *Biochim Biophys Acta* **1302**:93-109 (1996)). Like other members of this receptor superfamily, all of the PPAR isoforms contain a central DNA binding domain that recognizes response elements in the promoters of their target genes (see, for example, Latruffe et al. in *Biochimie* 79:81-94 (1997)). PPAR response elements (PPRE) are composed of a directly repeating core-site separated by 1 nucleotide (see Kliewer et al., in *Nature* **358**:771-4 (1992)). In order to recognize a PPRE, PPARs must heterodimerize with the 9-cis retinoic acid receptor (RXR).

The peroxisome proliferator activated receptors (PPARs) preferentially bind to DNA, i.e., response elements, as heterodimers with a common partner, the retinoid X (or 9-*cis* retinoic acid) receptor (RXR; see, for example, Kliewer et al., in *Nature* 355:446-449 (1992); Leid et al, in *Cell* 68:377-395 (1992); Marks et al., in *EMBO J.* 11:1419-1435 (1992); Zhang et al., in *Nature* 355:441-446 (1992); and Issemann et al., in *Biochimie.* 75:251-256 (1993). Once bound to a response element, PPARs activate transcription following binding of ligand to the C-terminal ligand binding domain thereof. Due to the key role of ligands for the activation of transcription, an intense search for the identification of ligands for members of the PPAR family has been undertaken by a number of research groups.

PPARα has been identified as a vertebrate nuclear hormone receptor which regulates genes involved in fatty acid (FA) degradation (δ- and ω-oxidation; see Schoonjans et al., in *Biochim Biophys Acta* **1302**:93-109 (1996)). PPARα is highly

expressed in the liver and was originally identified by Green and colleagues as a molecule that mediates the transcriptional effects of drugs that induce peroxisome proliferation in rodents (see Issemann & Green in *Nature* 347:645-50 (1990)). Mice lacking functional PPARα are incapable of responding to these agents and fail to induce expression of a variety of genes required for the metabolism of FAs in peroxisomes, mitochondria and other cellular compartments (see Lee et al., in *Mol Cell Biol* 15:3012-3022 (1995)). As a result, PPARα-deficient mice inappropriately accumulate lipid in response to pharmacologic stimuli.

PPARα appears to regulate FA oxidation, suggesting that PPARα ligands may represent endogenous signals for FA degradation (see Schoonjans et al., supra). Fatty acids (FAs) are ubiquitous biological molecules that are utilized as metabolic fuels, as covalent regulators of signaling molecules and as essential components of cellular membranes. It is thus logical that FA levels should be closely regulated. Indeed, some of the most common medical disorders in industrialized societies (e.g., cardiovascular disease, hyperlipidemia, obesity and insulin resistance) are characterized by altered levels of FAs or their metabolites (see, for example, Durrington, in *Postgrad Med J* 69 **Suppl 1**, S18-25; discussion S25-9 (1993) and Reaven, in *J Intern Med Suppl* 736:13-22 (1994)).

PPARγ is preferentially expressed in adipose tissue. PPARγ-activation leads to adipocyte differentiation and improved insulin signaling of mature adipocytes. 15-deoxy-$\Delta^{12,14}$-prostaglandin J_2 (15d-J_2) has been identified as a ligand for PPARγ (see, for example, Forman et al., in *Cell* 83:803-12 (1995) and Kliewer et al., in *Cell* 83:813-9 (1995)). Activation of PPARγ by 15d-J_2 or its synthetic analogs (e.g., thiazolidinediones; see Forman et al., supra) promotes differentiation of pre-adipocytes into mature, triglyceride-containing fat cells. Similarly, thiazolidinediones have been shown to increase body weight in animals (see, e.g., Zhang et al. (1996) *J Biol Chem* 271:9455-9459), suggesting that 15d-J_2 may be utilized as an *in vivo* signal to store fatty acids (FAs) in the form of triglycerides.

Accordingly, there is a need in the art for new agents and compositions which allow the modulation of hormone mediated processes. This and other needs in the art are addressed by the present invention.

BRIEF DESCRIPTION OF THE INVENTION

In accordance with the present invention, it has been discovered that retinoic acid receptor (RAR) antagonists are capable of modulating processes mediated by other members of the steroid/thyroid hormone receptor superfamily, including permissive receptors such as PPARs (e.g., PPARα, PPARδ and PPARγ). Indeed, it has been discovered that RAR antagonists, in combination with agonists for members of the steroid/thyroid hormone receptor superfamily, are capable of inducing and/or enhancing processes mediated by such members.

BRIEF DESCRIPTION OF THE FIGURES

Figure 1 illustrates the effects of AGN193109 on PPARγ response (induction of CD14 expression in myeloid cell lines), by comparing HL-60 (intact PPARγ response) and HL-60-CDM-1 cells (impaired PPARγ response, wherein no PPARγ is expressed).

Figure 2 illustrates the effects of the combination of the PPAR-γ agonist, PG-J2, the RXR agonist, LG 268, and the RAR antagonist, AGN 193109, on HL-60 cells.

Figure 3 provides several graphs summarizing the flow cytometry data for experiments depicted in Figure 2.

DETAILED DESCRIPTION OF THE INVENTION

In accordance with the present invention, there are provided compositions for modulating hormone mediated process(es) comprising:

at least one agonist for a member of the steroid/thyroid hormone receptor superfamily, and

optionally, at least one agonist for a heterodimer partner for said member; and

at least one antagonist for a second member of the steroid/thyroid hormone receptor superfamily, preferably for a non-permissive member such as RAR.

As employed herein, the phrase "members of the nuclear receptor superfamily" (also known as "members of the steroid/thyroid hormone superfamily of receptors" or "intracellular receptors") refers to hormone binding proteins that operate as ligand-dependent transcription factors, including identified members of the steroid/thyroid hormone superfamily of receptors for which specific ligands have not yet been identified (referred to hereinafter as "orphan receptors"). These hormone binding proteins have the intrinsic ability to bind to specific DNA sequences. Following binding, the transcriptional activity of target gene (i.e., a gene associated with the specific DNA sequence) is modulated as a function of the ligand bound to the receptor.

A member of the superfamily can be identified as a protein which contains the above-mentioned invariant amino acid residues, which are part of the DNA-binding domain of such known steroid receptors as the human glucocorticoid receptor (amino acids 421-486), the estrogen receptor (amino acids 185-250), the mineralocorticoid receptor (amino acids 603-668), the human retinoic acid receptor (amino acids 88-153), and the like. The highly conserved amino acids of the DNA-binding domain of members of the superfamily are as follows:

Cys - X - X - Cys - X - X - Asp* - X - Ala* - X - Gly* - X - Tyr* - X - X - X - X - Cys - X - X - Cys - Lys* - X - Phe - Phe - X - Arg* - X - X - X - X - X - X - X - X - (X - X -) Cys -X - X - X - X - X - (X - X - X -) Cys -X - X - X - Lys - X - X - Arg - X - X -Cys - X - X - Cys - Arg* - X - X - Lys* - Cys - X - X - X - Gly* - Met (SEQ ID NO:1);

wherein X designates non-conserved amino acids within the DNA-binding domain; the amino acid residues denoted with an asterisk are residues that are almost universally conserved, but for which variations have been found in some identified hormone receptors; and the residues enclosed in parenthesis are optional residues (thus, the DNA-binding domain is a minimum of 66 amino acids in length, but can contain several additional residues).

Members of the steroid/thyroid hormone superfamily of receptors (including the various isoforms thereof) include steroid receptors such as glucocorticoid receptor (GR), mineralocorticoid receptor (MR), estrogen receptor (ER), progesterone receptor (PR), androgen receptor (AR), vitamin D_3 receptor (VDR), and the like; plus retinoid receptors, such as the various isoforms of retinoic acid receptor (e.g., RARα, RARβ or RARγ), the various isoforms of retinoid X (or 9-*cis* retinoic acid) receptor (e.g., RXRα, RXRβ, or RXRγ), various isoforms of peroxisome proliferator-activated receptors (e.g., PPARα, PPARγ, PPARδ) and the like (see, e.g., U.S. Patent Nos. 4,981,784; 5,171,671; and 5,071,773); thyroid hormone receptor (T_3R), such as TRα, TRβ, and the like; steroid and xenobiotic receptor (SXR, see for example, Blumberg et al., *Genes Dev* (1998) **12(20)**:3195-205), RXR-interacting proteins (RIPs; see, e.g., Seol et al., *Mol Endocrinol* (1995) **9(1)**:72-85; Zavacki et al., *Proc Natl Acad Sci USA* (1997) **94(15)**:7909-14) including farnesoid X receptor (FXR; see for example, Forman et al., *Cell* (1995) **81(5)**:687-93), BXR (Blumberg et al., Genes Dev (1998) 12(9):1269-77), Hanley et al., *J Clin Invest* (1997) **100(3)**:705-12, O'Brien et al., *Carcinogenesis* (1996) **17(2)**:185-90), pregnenolone X receptor (PXR; see for example, Schuetz et al., *Mol Pharmacol* (1998) **54(6)**:1113-7), liver X receptor (LXR, see, e.g., Peet et al., *Curr Opin Genet Dev* (1998) **8(5)**:571-5), insect derived receptors such as the ecdysone receptor (EcR), the ultraspiracle receptor (see, for example, Oro

et al., in *Nature* **347**:298-301 (1990)), and the like; as well as other gene products which, by their structure and properties, are considered to be members of the superfamily, as defined hereinabove, including the various isoforms thereof (see, e.g., Laudet, V., *J Mol Endocrinol* (1997) **19(3)**:207-26).

In accordance with the present invention, the compositions for modulating hormone mediated process are capable of modulating the activity of complexes comprising homodimeric or heterodimeric member(s) of the steroid/thyroid hormone superfamily of receptors. It is readily recognized that a number of receptors preferentially bind to DNA as homodimers or heterodimers. Homodimeric members of the steroid/thyroid hormone receptor superfamily include GR, TR, RAR, RXR, and the like (see, e.g., Beato et al., *Steroids* **61(4)**:240-51 (1996)). Alternatively, RAR, VDR, TR, PPAR, SXR, OR1, SHP, LXR, BXR, and the like, preferentially form heterodimers with a common partner, e.g., RXR (see, for example, Dong et al., Biochemistry (1998) 27(30):10691-700; Yu et al., in *Cell* **67**:1251-1266 (1991); Bugge et al., in *EMBO J.* **11**:1409-18 (1992); Kliewer et al., in *Nature* **355**:446-449 (1992); Leid et al, in *Cell* **68**:377-395 (1992); Marks et al., in *EMBO J.* **11**:1419-1435 (1992); Zhang et al., in *Nature* **355**:441-446 (1992); and Issemann et al., in *Biochimie.* **75**:251-256 (1993). Similarly, other receptors, e.g., EcR, will form heterodimers with the RXR homolog, ultraspiracle. In a preferred embodiment of the present invention, the invention compositions will modulate the activity of permissive heterodimers. Permissive heterodimeric members of the steroid/thyroid hormone receptor superfamily are well known to those skilled in the art and include PPAR:RXR, LXR:RXR, NGFI-B:RXR, NURR1:RXR, FXR:RXR, BXR:RXR, SXR:RXR, and the like.

As employed herein, the term "agonist (or agonist precursor) for a member of the steroid/thyroid hormone superfamily of receptors" (i.e., intracellular receptor) refers to a substance or compound which, in its unmodified form (or after conversion to its "active" form), inside a cell, binds to receptor protein, thereby creating an

agonist/receptor complex, which in turn can activate an appropriate hormone response element. An agonist therefore is a compound which acts to modulate gene transcription for a gene maintained under the control of a hormone response element, and includes compounds such as hormones, growth substances, non-hormone compounds that modulate growth, and the like. Agonists include steroid or steroid-like hormone, retinoids, thyroid hormones, pharmaceutically active compounds, and the like. Individual agonists may have the ability to bind to multiple receptors. Preferably, the agonist is for a member which forms a heterodimer (or homodimer) with additional or other members. In a more preferred embodiment, the agonist for a member of the steroid/thyroid hormone superfamily of receptors is an agonist for a permissive receptor, such as PPAR, LXR, RXR interacting proteins (RIPs including, for example, FXR), and the like. In the most preferred embodiment, the agonist is for a member other than retinoic acid receptor.

Agonists for individual members of the steroid/thyroid hormone superfamily of receptor are well known in the art. For example, agonists for retinoids are described in *The Retinoids : Biology, Chemistry, and Medicine* (Sporn, Roberts & Goodman, eds. (Raven Press, 1993), the entire contents of which is hereby incorporated by reference herein).

Peroxisome proliferator-activated receptor (PPAR) agonist(s) contemplated for use herein are well known in the art. See, for example, Peroxisome Proliferators: Unique Inducers of Drug-Metabolizing Enzymes (Pharmacology and Toxicology) David E. Moody (Editor) (July 1994), the entire contents of which are hereby incorporated by reference herein. As readily recognized by those of skill in the art, a variety of PPAR agonists, both synthetic and naturally occurring, can be used in accordance with the present invention. Exemplary PPAR agonists include hypolipidemic drugs, polyunsaturated fatty acids, eicosanoids, thiazolidines (Komers et al., Physiol Res (1998) 47(4):215-25), benzene compounds, anti-inflammatory compounds (NSAIDs), and the like (see, e.g., Tajima et al., WO9915520, Collins et

al., *J Med Chem* **41(25)**:5037-54 (1998), Forman et al., Proc Natl Acad Sci U S A (1997) 94(9):4312-7, Willson et al., Curr Opin Chem Biol (1997) 1(2):235-41, Sorensen et al., Vitam Horm (1998) 54:121-66, Berger et al., J Biol Chem (1999) 274(10):6718-25, Wilson et al., Ann N Y Acad Sci (1996) 804:276-83). Preferred PPAR agonists include troglitazone, WY14,643, GW0072, rosiglitazone (BRL 49653), L-764406, 15-deoxy-Delta12, 14-prostaglandin J2 (15d-PGJ2) and oxidized linoleic acid (9- and 13-HODE), (2S)-((2-benzoylphenyl) amino)-3-4-[2-(5-methyl-2-phenyloxazol-4-yl) ethoxy] phenylpropanoic acid, 2(S)-((2-benzoylphenyl)amino)-3,4- [2-(5-methyl-2-pyridin-4-yloxazol-4-yl) ethoxy] phenylpropionic acid, 2(S)-((2-benzoylphenyl)amino)-3-(4-2- [5-methyl-2-(4-methylpiperazin-1-yl)thiazol-4-yl]ethoxyphenyl) propionic acid, (2S)-3-(4-(benzyloxy)phenyl)-2-((1-methyl-3-oxo-3-phenylpropenyl)amino) propionic acid, (2S)-((2-benzoylphenyl)amino)-3-4- [2-(methylpyridin-2-ylamino) ethoxy]phenylpropionic acid, 3-4-[2-(benzoxazol-2-ylmethylamino) ethoxy] phenyl-(2S)-((2- benzoylphenyl)amino)propanoic acid, and the like.

Additional agonists contemplated for use in the practice of the present invention depend on the target receptor and are known to those skilled in the art, including benzoate metabolites for BXR (e.g., Blumberg et al., *Genes Dev.* (1998) **12(9)**:1269-77), farnesoids and bile acids for FXR (e.g., Parks et al., *Science* (1999) **284(5418)**:1365-8), oxysterols for LXR (e.g., Janowski et al., *Proc Natl Acad Sci U S A* (1999) **96(1)**:266-71), and the like.

The invention composition further optionally comprises at least one agonist for a second member of the steroid/thyroid hormone superfamily of receptors, i.e., a heterodimer partner. Those of skill in the art readily recognize those members which can form heterodimer partners, including RXR, ultraspiracle NGFI-B, NURR1, and the like. RXR agonist(s) contemplated for use herein are well known in the art. See, for example, The Retinoids, supra. As readily recognized by those of skill in the art, a variety of RXR agonists, both synthetic and naturally occuring, can be used in

accordance with the present invention. Examplary RXR agonists include 3-substituted (tetramethyltetrahydronaphthyl)carbonylbenzoic acids (Canan Koch et al., J Med Chem (1999) 42(4):742-50), (E,E,E)-7-(1,2,3,4-tetrahydroquinolin-6-yl)-7-alkyl-6-fluoro-3-methylhepta-2, 4, 6-trienoic acid derivatives (Hibi et al., J Med Chem (1998) 41(17):3245-52), diaryl sulfide retinoid analogs (Beard et al., J Med Chem (1996) 39(18):3556-63), phytanic acid (Lemotte et al., Eur J Biochem (1996) 236(1):328-33), tricyclic compounds (U.S. Patents Nos. 5,770,383, 5,770,382 and 5,770,378), trienic compounds (U.S. Patent No. 5,721,103), and the like (see also, Jiang et al., Biochem Pharmacol (1995) 50(5):669-76), Heyman WO9710819). Preferred RXR agonists include LGD1069 (Bischoff et al., Cancer Res (1998) 58(3):479-84), LG100153, E(-2-[2-(5,6,7,8-Tetrahydro-3,5,5,8,8-pentamethyl-2-naphthyl) propen-1-yl]-4-thiophenecarboxylic acid (AGN 191701), 2-(5,6,7,8-Tetrahydro-5,5,8,8-tetramethyl-2-naphthyl)-2-(4-carboxylphenyl)-1,3-dioxolane (SR11237), Standeven et al., Biochem Pharmacol (1997) 54(4):517-24), HX600 or HX630, and the like.

In accordance with the present invention, the invention compositions will comprise at least one antagonist for members of the steroid/thyroid hormone superfamily of receptors, including antagonists for other non-permissive receptors, such as antagonists for RAR, TR, VDR, and the like. Preferably, the antagonist will be an RAR antagonist. Those of skill in the art will readily recognize antagonists which can be employed in the practice of the present invention. As readily recognized by those of skill in the art, a variety of retinoic acid receptor (RAR) antagonists, both synthetic and naturally occuring, can be used in accordance with the present invention. Examplary RAR antagonists include dicarba-closo-dodecaboranes (Iijima et al., Chem Pharm Bull (Tokyo) (1999) 47(3):398-404), hydroanthracenyl, benzochromenyl and benzothiochromenyl retinoids (Vuligonda et al., Bioorg Med Chem Lett (1999) 9(5):743-8), diarylacetylenes, benzoic acid derivatives (see, e.g., Kagechika, H. (1994) *Yakugaku Zasshi* **114(11)**:847-862; Eckhardt et al. (1994) *Toxicol Lett* **70(3)**:299-308; Yoshimura et al. (1995) *J Med Chem* **38(16)**:3163-3173; Chen et al. (1995) *EMBO* **14(6)**:1187-1197; Teng et al. (1997) *J Med Chem*

40(16):2445-2451); naphthalenyl analogs (see, e.g., Johnson et al. (1995) *J Med Chem* **38(24)**:4764-4767; Agarwal et al. *J Biol Chem* **271(21)**:12209-12212: Umemiya et al. (1996) *Yakugaku Zasshi* **116(12)**:928-941); aryl-substituted and aryl and (3-oxo-1-propenly)-substituted benzopyran, benzothiopyran, 1,2-dihydroquinoline, and 5,6-dihydronaphthalene derivatives (Klein et al. U.S. Pat. Nos. 5,877,207 and 5,776,699), adamantyl-substituted biaromatic compounds (Bernardon and Charpentier, U.S. Pat. No. 5,877,342), 1-phenyl-adamantane derivatives (Bernardon and Bernardon EP 776885), polyaromatic heterocyclic compounds (Charpentier et al. U.S. Pat. No. 5,849,798), dihydronaphthalene derivatives (Beard et al., U.S. Pat. No. 5,808,124 and Johnson et al. U.S. Pat. No. 5,773,594), 4-phenyl (benozopyranoyl or naphthoyl) amidobenzoic acid derivatives (Chandraratna et al. WO 98/US/13065), diazepinylbenzoic acid derivatives (Umemiya et al., J Med Chem (1997) 40(26):4222-34), tetrahydronaphthalene derivatives (Vuligonda et al. U.S. Pat. No. 5,763,635, 5,741,896 and 5,723,666), aryl-and heteroarylcyclohexenyl substituted alkenes (Beard et al. U.S. Patent No. 5,760,276), dibenzofuran compounds, including aromatic dibenzofuran compounds (Charpentier et al. U.S. Pat. No. 5,702,710, Charpentier and Bernard U.S. Pat. No. 5,747,530), N-aryl substituted tetrahydroquinolines (Beard et al. U.S. Pat. No. 5,739,338), benzo[1,2-g]-chrom-3-ene and benzo[1,2-g]-thiochrom-3-ene derivatives (Vuligonda et al. U.S. Pat. No. 5,728,846), and the like (see also, Chandraratna, RA, Cutis (1998) 61(2 Suppl):40-5).

Examples of specific RAR antagonists contemplated for use herein include LE135 (Umemiya et al. (1996) *Yakugaku Zasshi* **116(12)**:928-941), LE511, LE540, LE550 (Li et al., *J Biol Chem* (1999) **274(22)**:15360-6; Umemiya et al. (1996) *Yakugaku Zasshi* **116(12)**:928-941), Ro41-5253 (Keidel et al. (1994) *Mol Cell Biol* **14(1)**:287-298), SR11330, SR11334, SR11335 (Lee et al. (1996) *J. Biol Chem* **271(20)**:11897-11903), BMS453, BMS411 (Chen et al. (1995) *EMBO* **14(6)**:1187-1197), CD2366 and CD2665 (Meister et al., *Anticancer Res.* (1998) **18(3A)**:1777-1786), ER27191 (Uemo et al., *Leuk. Res.* (1998) **22(6)**:517-525), AGN 193109

(Johnson et al., *Bioorg Med Chem Lett* (1999) 9(4):573-6), 4-[4,5,7,8,9,10-hexahydro-7,7,10,-10-tetramethyl-1-(3-pyridylmethyl)anthra[1,2-b]pyrrol-3-yl]benzoic acid, 4-[4,5,7,8,9,10-hexahydro-7,7,10,10-tetramethyl-1-(3-pyridylmethyl)-5-thiaanthral[1,2-b]pyrrol-3-yl]benzoic acid, 4-[4,5,7,8,9,10-hexahydro-7,7,10,10-tetramethyl-1-(3-pyridylmethyl)anthra[2,1-d]pyrazol-3-yl]benzoic acid (Yoshimura et al. (1995) *J Med Chem* **38(16)**:3163-3173), AGN193109 (Agarwal et al. *J Biol Chem* **271(21)**:12209-12212), and the like. A presently preferred class of RAR antagonists contemplated for use according to the invention are aryl dihydronaphthalenyl derivatives of acetylene. An especially preferred RAR antagonist contemplated for use herein is 4-[[5,6-dihydro-5,5-dimethyl-8-(4-methylphenyl)-2-naphthalenyl]ethynyl] benzoic acid.

In accordance with another aspect of the present invention, there are provided methods for modulating hormone mediated processes, the methods comprising introducing an effective amount of at least one retinoic acid receptor (RAR) antagonist in combination with at least one agonist for the member into the system. Preferably, the agonist is an agonist for a member of the steroid/thyroid hormone superfamily of receptors which form heterodimers or homodimers with other or additional members. More preferably, the agonist is for a permissive receptor. In the presently most prefered embodiment, the agonist is not for retinoic acid receptor.

In yet another preferred embodiment of the present invention, there are provided methods for relieving, in a biological system, the inhibition of hormone mediated process(es), such as process(es) mediated by peroxisome proliferator activated receptor(s) (PPARs). In a presently preferred aspect, the invention method relieves inhibition of hormone mediated processes caused by retinoic acid receptor (RAR) and agonist(s) thereof. Alternatively, or in addition, there are provided methods for inducing hormone mediated process(es) in a biological system by peroxisome proliferator activated receptor(s) (PPARs). Invention methods comprise introducing an effective amount of at least one retinoic acid receptor (RAR) antagonist, alone, or in combination with PPAR agonists, into said system.

In accordance with yet another embodiment of the present invention, the invention method further comprises administering or co-administering an effective amount of at least one agonist for a heterodimer partner for the member, e.g., RXR, NURR1, ultraspiracle, and the like.

As employed herein, the term "modulate" refers to the ability of compound(s) to either directly (by binding to the receptor as a ligand) or indirectly (by relieving the inhibition of process(es) mediated by members of the steroid/thyroid hormone receptor superfamily, or as a precursor and/or facilitator for a ligand or an inducer which promotes production of ligand from a precursor, or combinations thereof) induce and/or enhance expression of gene(s) maintained under hormone expression control, or to repress expression of gene(s) maintained under such control.

As employed herein, the phrase "relieving ... the inhibition of process(es) ..." refers to the ability of a suitable compound, e.g., RAR antagonist, to either directly (by binding to the receptor as a ligand) or indirectly (as a precursor for a ligand or an inducer which promotes production of ligand from a precursor) counteract or divert the ability of members of the steroid/thyroid hormone receptor superfamily, and agonists thereof, to inhibit or interfere with the expression of gene(s) maintained under hormone expression control. Preferably, members of the steroid/thyroid hormone receptor superfamily contemplated for use in the practice of the present invention include non-permissive receptors such as retinoic acid receptor.

As employed herein, the term "inducing" refers to the ability of a modulator for a member of the steroid/thyroid hormone receptor superfamily to either directly (by binding to the receptor as a ligand) or indirectly (as a precursor for a ligand or an inducer which promotes production of ligand from a precursor) promote expression of gene(s) maintained under hormone expression control.

As employed herein, the phrase "biological system" refers to an intact organism or a cell-based system containing the various components required for response to the ligands described herein, e.g., a member of the steroid/thyroid hormone receptor superfamily, a heterodimer partner for the member (e.g., RXR), and a reporter responsive to the heterodimer (which typically comprises a response element (RE) in operative communication with a reporter gene; suitable reporters include luciferase, chloramphenicol transferase, β-galactosidase, and the like).

As employed herein, the phrase "hormone mediated processes" refers to biological, physiological, endocrinological, and other bodily processes which are mediated by receptor or receptor combinations which are responsive to the ligands described herein. Modulation of such processes can be accomplished *in vitro* or *in vivo*. *In vivo* modulation can be carried out in a wide range of subjects, such as, for example, humans, rodents, sheep, pigs, cows, and the like. Exemplary processes contemplated to be modulated include neoplastic diseases, inflammatory or infectious diseases, and the like.

As employed herein, the phrase "process(es) mediated by peroxisome proliferator activated receptor(s) (PPARs)" refers to processes which are manifested by expression or repression of expression of PPAR-responsive genes. Thus, for example, "PPARα-responsive genes" refers to genes whose expression products are involved in the biological, physiological, endocrinological, and other bodily processes which are mediated by receptor or receptor combinations which are responsive to the PPARα ligands described herein (e.g., genes involved in fatty acid metabolism in peroxisomes, mitochondria and other cellular compartments (including FA degradation (by β- and ω-oxidation), and the like). Modulation of such processes can be accomplished *in vitro* or *in vivo*. *In vivo* modulation can be carried out in a wide range of subjects, such as, for example, humans, rodents, sheep, pigs, cows, and the like.

As employed herein, the phrase "PPARδ-responsive genes" refers to genes whose expression products are involved in the biological, physiological, endocrinological, and other bodily processes which are mediated by receptor or receptor combinations which are responsive to PPARδ ligands. Modulation of such processes can be accomplished *in vitro* or *in vivo*. *In vivo* modulation can be carried out in a wide range of subjects, such as, for example, humans, rodents, sheep, pigs, cows, and the like.

As employed herein, the phrase "PPARγ-responsive genes" refers to genes whose expression products are involved in the biological, physiological, endocrinological, and other bodily processes which are mediated by receptor or receptor combinations which are responsive to PPAR-γ ligands (e.g., cell differentiation to produce lipid-accumulating cells, regulation of insulin-sensitivity and blood glucose levels, especially as related to hypoglycemia/hyperinsulinism (resulting, for example, from abnormal pancreatic beta-cell function, insulin-secreting tumors and/or autoimmune hypoglycemia due to autoantibodies to insulin, the insulin receptor or autoantibodies that are stimulatory to pancreatic beta-cells), the formation of macrophages which lead to the development of atherosclerotic plaques, and the like). Modulation of such processes can be accomplished *in vitro* or *in vivo*. *In vivo* modulation can be carried out in a wide range of subjects, such as, for example, humans, rodents, sheep, pigs, cows, and the like.

As employed herein, the phrase "effective amount" refers to levels of compound sufficient to provide circulating concentrations high enough to modulate the expression of an isoform of PPAR. Such a concentration typically falls in the range of about 10 nM up to 2 mM; with concentrations in the range of about 100 nM up to 500 nM being preferred. Since the activity of different compounds described herein may vary considerably, and since individual subjects may present a wide variation in severity of symptoms, it is up to the practitioner to determine a subject's response to treatment and vary the dosages accordingly.

The above-described biologically active compounds can be administered in a variety of forms (e.g., in combination with a pharmaceutically acceptable carrier therefor) and by a variety of modes of delivery. Exemplary pharmaceutically acceptable carriers include carriers suitable for oral, intravenous, subcutaneous, intramuscular, intracutaneous, and the like administration. Administration in the form of creams, lotions, tablets, dispersible powders, granules, syrups, elixirs, sterile aqueous or non-aqueous solutions, suspensions or emulsions, and the like, is contemplated.

For the preparation of oral liquids, suitable carriers include emulsions, solutions, suspensions, syrups, and the like, optionally containing additives such as wetting agents, emulsifying and suspending agents, sweetening, flavoring and perfuming agents, and the like.

For the preparation of fluids for parenteral administration, suitable carriers include sterile aqueous or non-aqueous solutions, suspensions, or emulsions. Examples of non-aqueous solvents or vehicles are propylene glycol, polyethylene glycol, vegetable oils, such as olive oil and corn oil, gelatin, and injectable organic esters such as ethyl oleate. Such dosage forms may also contain adjuvants such as preserving, wetting, emulsifying, and dispersing agents. They may be sterilized, for example, by filtration through a bacteria-retaining filter, by incorporating sterilizing agents into the compositions, by irradiating the compositions, or by heating the compositions. They can also be manufactured in the form of sterile water, or some other sterile injectable medium immediately before use.

As used herein, co-administration of two pharmacologically active compounds refers to the delivery of two separate chemical entities, whether *in vitro* or *in vivo*. Co-administration refers to the simultaneous delivery of separate agents; to the simultaneous delivery of a mixture of agents; as well as to the delivery of one agent

followed by delivery of the second agent. In all cases, agents that are co-administered are intended to work in conjunction with each other.

Each of the references and U.S. and foreign patents cited herein are hereby incorporated by reference in its entirety. The invention will now be described in greater detail by reference to the following non-limiting examples.

Example 1

Cell Culture and Transfection

CV-1 cells were grown and transfected as described by Forman et al., in *Cell* 83:803-12 (1995). The reporter construct, PPREx3 TK-LUC, contained 3 copies of the acyl CoA oxidase PPRE upstream of the Herpes virus thymidine kinase promoter (see Kliewer et al., in *Nature* 358:771-4 (1992)). Expression vectors contained the cytomegalovirus IE promoter/enhancer (pCMX) upstream of wild-type mouse PPARα, mouse PPARγ1-DN (Met^{105}-Tyr^{475}), mouse PPARα*-DN (Leu^{69}-Tyr^{440}), mouse PPARα-G (Glu^{282}-> Gly) (see Hsu et al., in *Mol Pharmacol* 48:559-67 (1995)) or E. coli ß-galactosidase as an internal control. Cells were exposed to the compounds for 24 hours then harvested and assayed for luciferase and β-galactosidase activity. All points were performed in triplicate and varied by less than 10%. Normalized luciferase activity was determined and plotted as fold-activation relative to untreated cells. Each experiment was repeated three or more times with similar results.

Example 2

Electrophoretic Mobility Shift Assays

In vitro translated mouse PPARα (0.2 ml) and human RXRα (0.1 ml) were incubated for 30 minutes at room temperature with 100,000 cpm of Klenow-labeled acyl CoA oxidase PPRE as described by Forman et al., in *Cell* 81:687-93 (1995), but with 150 mM KCl.

Example 3

Activation of PPARα by Diarylacetylenes

In order to evaluate the ability of RAR antagonists to relieve the inhibition of processes mediated by PPARs, CV-1 cells are transiently transfected with a PPAR responsive reporter, PPAR expression vectors and then treated with 4-[[5,6-dihydro-5,5-dimethyl-8-(4-methylphenyl)-2-naphthalenyl]ethynyl] benzoic acid. Wy 14,643 and rosiglitazone (BRL 49653) are included as positive controls since these compounds selectively activate PPAR α and γ, respectively (see Forman et al., in *Cell* **83**:803-12 (1995), Kliewer et al., in *Cell* **83**:813-9 (1995) and Kliewer et al., in *Proc Natl Acad Sci USA* **91**:7355-9 (1994)).

4-[[5,6-dihydro-5,5-dimethyl-8-(4-methylphenyl)-2-naphthalenyl]ethynyl] benzoic acid is found to activate PPARα maximally at about 300 mM.

Example 4

Activation of PPARα by Benzoic Acid Derivatives

In order to evaluate the ability of RAR antagonists to relieve the inhibition of processes mediated by PPARs, CV-1 cells are transiently transfected with a PPAR responsive reporter, PPAR expression vectors and then treated with 4-[4-(2,2,2,-trifluoro-1-methoxyethyl)5,6,7,8-tetrahydro-5,5,8,8-tetramethyl-2-anthracenyl]benzoic acid (SR11335; Lee et al. *J Biol Chem* **271**:11897-11903). Wy 14,643 and BRL 49653 are included as positive controls since these compounds selectively activate PPAR α and γ, respectively (see Forman et al., in *Cell* **83**:803-12 (1995), Kliewer et al., in *Cell* **83**:813-9 (1995) and Kliewer et al., in *Proc Natl Acad Sci USA* **91**:7355-9 (1994)).

4-[4-(2,2,2,-trifluoro-1-methoxyethyl)5,6,7,8-tetrahydro-5,5,8,8-tetramethyl-2-anthracenyl]benzoic acid is found to activate PPARα maximally at about 300 mM.

Example 5

Retinoid Antagonists Relieve the Retinoic Acid Induced Inhibition of PPAR Mediated Processes

To confirm the effect of an RAR antagonist on PPARα/RXRα, studies are done with the homologous malic enzyme gene promoter (pMECAT -490/+31; see Castelein et al. (1994) *J Biol Chem* **269**:26754-26758). The -490/+31 malic enzyme promoter sequence is inserted in a pOCAT2 reporter and used in this transfection experiment with receptor plasmids in COS cells. Increasing amounts of an RAR agonist (e.g., retinoic acid) is presented with a constant amount of PPARα/RARα expressing vectors. The cells are treated with either DMSO, ciprofibrate (100 mM) or with the combination of ciprofibrate and an RAR antagonist.

The activity of the pOCAT2 reporter is reduced alone in the presence of an RAR agonist. PPARα/RXRα stimulate CAT activity of the pMECAT reporter to almost the same extent, regardless of the absence or presence of their respective ligands. RAR agonists suppress CAT activation by the non-liganded PPARα/RARα in a dose dependent manner, and, to a lesser extent, also suppress CAT activation by the ciprofibrate activated receptors. However, RAR agonist is completely unable to counteract PPARα/RARα activated by ciprofibrate and an RAR antagonist. These results show that RAR antagonists may relieve the repression of PPARα transactivation caused by RAR agonist.

Example 6

RAR antagonists enhances PPARγ mediated biological processes

A myeloid cell line HL-60 was treated with the PPAR gamma activator, PG-J2 (5μM), the RXR activator, LG 268 (100nM), the RAR/RXR activator, 9-cis retinoic acid (100 nM), and the RAR antagonist AGN 193109 (100nM), individually or in different combinations. The expression levels of monocytic cell surface marker CD14 is measured over two days. As a comparison, HL-60-CDM-1 cells which do not express PPARγ and thus have impaired PPARγ response, were also treated with the above ligands, individually or in various combinations (Figure 1).

Figure 1 and 2 illustrate the dramatic effect of the combination of PPAR-γ ligand, PG-J2, the RXR agonist, LG268, and the RAR antagonist, AGN193109, on HL-60 cells.

While the invention has been described in detail with reference to certain preferred embodiments thereof, it will be understood that modifications and variations are within the spirit and scope of that which is described and claimed.

That which is claimed is:

1 A composition for modulating hormone mediated process(es) comprising:

at least one agonist for a member of the steroid/thyroid hormone receptor superfamily,

at least one RAR antagonist, and

optionally, at least one agonist for a heterodimer partner for said member.

2. A composition according to claim 1, wherein said member of the steroid/thyroid hormone receptor superfamily is a permissive receptor.

3. A composition according to claim 2, wherein said permissive receptor is PPAR, NGFI-B, NURR1, FAR or LXR.

4. A composition according to claim 3, wherein said agonist for said permissive receptor is a PPAR agonist.

5. A composition according to claim 4, wherein said PPAR agonist is a hypolipidemic drug, a polyunsaturated fatty acid, an eicosanoid, a thiazolidine, a benzene compound, an anti-inflammatory compound (NSAID) , or mixtures thereof.

6. A composition according to claim 5, wherein said PPAR agonist is troglitazone, WY14,643, GW0072, rosiglitazone (BRL 49653), L-764406, 15-deoxy-Delta12, 14-prostaglandin J2 (15d-PGJ2) and oxidized linoleic acid (9- and 13-HODE), (2S)-((2-benzoylphenyl) amino)-3-4-[2-(5-methyl-2-phenyloxazol-4-yl) ethoxy] phenylpropanoic acid, 2(S)-((2-benzoylphenyl)amino)-3,4-[2-(5-methyl-2-pyridin-4-yloxazol-4-yl) ethoxy] phenylpropionic acid, 2(S)-((2-benzoylphenyl)amino)-3-(4-2- [5-methyl-2-

(4-methylpiperazin-1-yl)thiazol-4-yl]ethoxyphenyl) propionic acid, (2S)-3-(4-(benzyloxy)phenyl)-2-((1-methyl-3-oxo-3-phenylpropenyl)amino) propionic acid, (2S)-((2-benzoylphenyl)amino)-3-4- [2-(methylpyridin-2-ylamino) ethoxy]phenylpropionic acid, 3-4-[2-(benzoxazol-2-ylmethylamino) ethoxy] phenyl-(2S)-((2- benzoylphenyl)amino)propanoic acid, or mixtures thereof.

7. A method according to claim 4 wherein said process is mediated by PPAR-α.

8. A method according to claim 4 wherein said process is mediated by PPAR-δ.

9. A method according to claim 4 wherein said process is mediated by PPAR-γ.

10. A composition according to claim 1, wherein said heterodimer partner is RXR or ultraspiracle.

11. A composition according to claim 10, wherein said agonist for said heterodimer partner is an RXR agonist.

12. A composition according to claim 11, wherein said RXR agonist is a 3-substituted (tetramethyltetrahydronaphthyl)carbonylbenzoic acid, an (E,E,E)-7-(1,2,3,4-tetrahydroquinolin-6-yl)-7-alkyl-6-fluoro-3-methylhepta-2, 4, 6-trienoic acid derivative, a diaryl sulfide retinoid analog, phytanic acid, a tricyclic or trienic compound, or mixtures thereof.

13. A composition according to claim 1, wherein said RAR antagonist is a dicarba-closo-dodecaborane, a hydroanthracenyl, a benzochromenyl and/or a benzothiochromenyl retinoid, a diarylacetylene, a benzoic acid derivative; naphthalenyl analog, an aryl-substituted, aryl and/or (3-oxo-1-propenly)-substituted benzopyran, a benzothiopyran, a 1,2-dihydroquinoline, a 5,6-dihydronaphthalene derivatives, an adamantyl-substituted biaromatic compound, a 1-phenyl-adamantane derivative, a polyaromatic heterocyclic compound, a dihydronaphthalene derivative, a 4-phenyl (benozopyranoyl or naphthoyl) amidobenzoic acid derivative, a diazepinylbenzoic acid derivative, a tetrahydronaphthalene derivative, an aryl- or heteroarylcyclohexenyl substituted alkene, a dibenzofuran compound, a N-aryl substituted tetrahydroquinolines, a benzo[1,2-g]-chrom-3-ene or benzo[1,2-g]-thiochrom-3-ene derivative, an aryl dihydronaphthalenyl derivative of acetylene, or mixtures thereof.

14. A composition according to claim 1, wherein said RAR antagonist is LE135, LE511, LE540, LE550, Ro41-5253, SR11330, SR11334, SR11335, BMS453, BMS411, CD2366, CD2665, ER27191, AGN193109, 4-[4,5,7,8,9,10-hexahydro-7,7,10,-10-tetramethyl-1-(3-pyridylmethyl)anthra[1,2-b]pyrrol-3-yl]benzoic acid, 4-[4,5,7,8,9,10-hexahydro-7,7,10,10-tetramethyl-1-(3-pyridylmethyl)-5-thiaanthral[1,2-b]pyrrol-3-yl]benzoic acid, 4-[4,5,7,8,9,10-hexahydro-7,7,10,10-tetramethyl-1-(3-pyridylmethyl)anthra[2,1-d]pyrazol-3-yl]benzoic acid, AGN193109, 4-[[5,6-dihydro-5,5-dimethyl-8-(4-methylphenyl)-2-naphthalenyl]ethynyl] benzoic acid, or mixtures thereof.

15. A method for modulating hormone mediated process(es) in a biological system, said method comprising introducing an effective amount of at least one retinoic acid receptor (RAR) antagonist into said system in combination with an effective amount of at least one agonist for a member of the steroid/thyroid hormone superfamily of receptors, wherein said agonist is not an agonist for retinoic acid receptor.

16. A method according to claim 15, wherein said method relieves the inhibition of retinoic acid receptor on process(es) mediated by members of the steroid/thyroid hormone superfamily of receptors.

17. A method according to claim 15, wherein said method enhances or induces process(es) mediated by members of the steroid/thyroid hormone superfamily of receptors.

18. A method according to claim 15 wherein said RAR antagonist is a dicarba-closo-dodecaborane, a hydroanthracenyl, a benzochromenyl and/or a benzothiochromenyl retinoid, a diarylacetylene, a benzoic acid derivative; naphthalenyl analog, an aryl-substituted, aryl and/or (3-oxo-1-propenly)-substituted benzopyran, a benzothiopyran, a 1,2-dihydroquinoline, a 5,6-dihydronaphthalene derivatives, an adamantyl-substituted biaromatic compound, a 1-phenyl-adamantane derivative, a polyaromatic heterocyclic compound, a dihydronaphthalene derivative, a 4-phenyl (benozopyranoyl or naphthoyl) amidobenzoic acid derivative, a diazepinylbenzoic acid derivative, a tetrahydronaphthalene derivative, an aryl- or heteroarylcyclohexenyl substituted alkene, a dibenzofuran compound, a N-aryl substituted tetrahydroquinolines, a benzo[1,2-g]-chrom-3-ene or benzo[1,2-g]-thiochrom-3-ene derivative, an aryl dihydronaphthalenyl derivative of acetylene, or mixtures thereof.

19. A method according to claim 18 wherein said RAR antagonist is an aryl dihydronaphthalenyl derivative of acetylene.

20. A method according to claim 15 wherein said RAR antagonist is LE135, LE511, LE540, LE550, Ro41-5253, SR11330, SR11334, SR11335, BMS453, BMS411, CD2366, CD2665, ER27191, AGN193109, 4-[4,5,7,8,9,10-hexahydro-7,7,10,-10-tetramethyl-1-(3-pyridylmethyl)anthra[1,2-b]pyrrol-3-yl]benzoic acid, 4-[4,5,7,8,9,10-hexahydro-7,7,10,10-tetramethyl-1-(3-pyridylmethyl)-5-thiaanthral[1,2-b]pyrrol-3-yl]benzoic acid, 4-[4,5,7,8,9,10-hexahydro-7,7,10,10-tetramethyl-1-(3-pyridylmethyl)anthra[2,1-d]pyrazol-3-yl]benzoic acid, AGN193109, 4-[[5,6-dihydro-5,5-dimethyl-8-(4-methylphenyl)-2-naphthalenyl]ethynyl] benzoic acid, or mixtures thereof.

21. A method according to claim 15, wherein said agonist is an agonist for a permissive receptor.

22. A method according to claim 21, wherein said agonist is a PPAR agonist.

23. A method according to claim 22, wherein said PPAR agonist is a hypolipidemic drug, a polyunsaturated fatty acid, an eicosanoid, a thiazolidine, a benzene compound, an anti-inflammatory compound (NSAID), or mixtures thereof.

24. A method according to claim 15 wherein said process is mediated by PPAR-α.

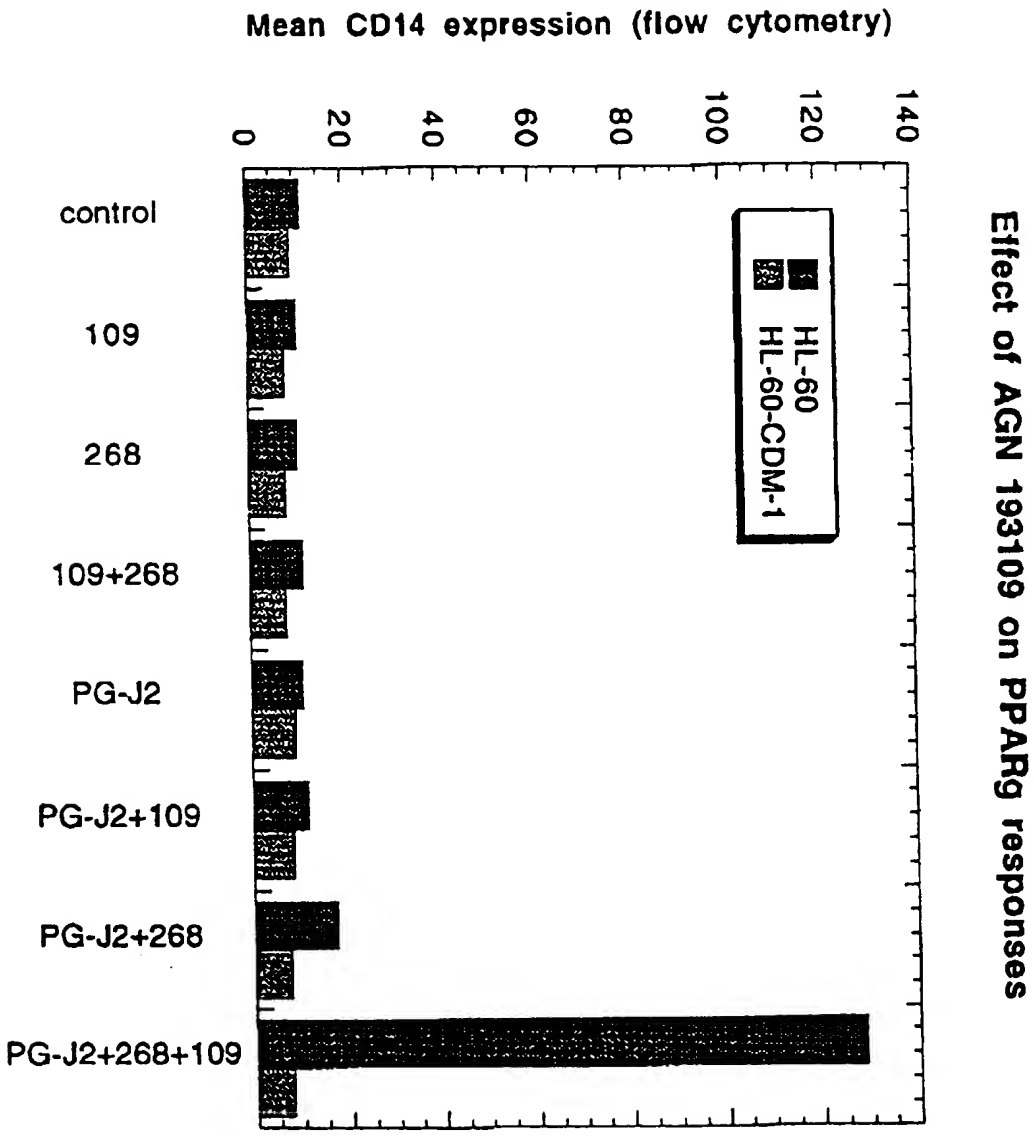
25. A method according to claim 15 wherein said process is mediated by PPAR-δ.

26. A method according to claim 15 wherein said process is mediated by PPAR-γ.

27. A method according to claim 15, wherein said method further comprises introducing an effective amount of at least one agonist for a heterodimer partner for said member.

28. A method according to claim 27, wherein said agonist for said heterodimer partner is an RXR agonist.

29. A method according to claim 28, wherein said RXR agonist is a 3-substituted (tetramethyltetrahydronaphthyl)carbonylbenzoic acid, a (E,E,E)-7-(1,2,3,4-tetrahydroquinolin-6-yl)-7-alkyl-6-fluoro-3-methylhepta-2, 4, 6-trienoic acid derivative, a diaryl sulfide retinoid analog, phytanic acid, a tricyclic or trienic compound, or mixtures thereof.


Mean CD14 expression (flow cytometry)
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20
40
60
80
100
120
140
control
109
268
109+268
PG-J2
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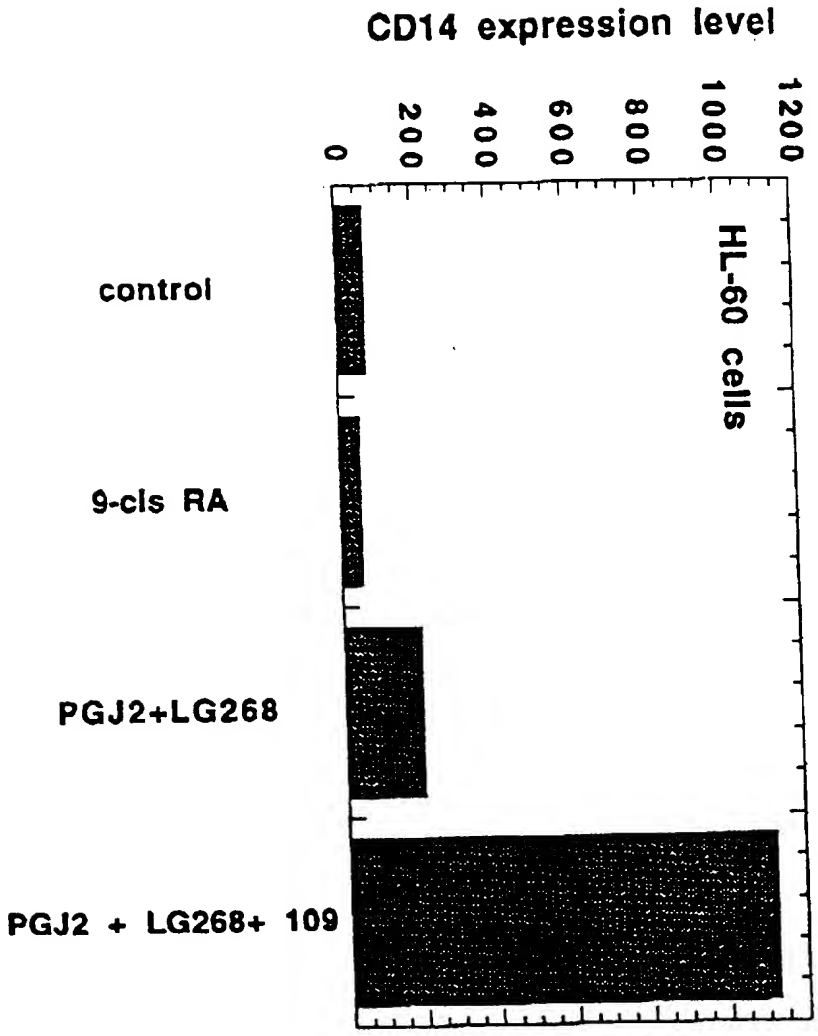
PG-J2+268
PG-J2+268+109
HL-60
HL-60-CDM-1
Effect of AGN 193109 on PPARg responses

FIG. 1


CD14 expression level
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800
1000
1200
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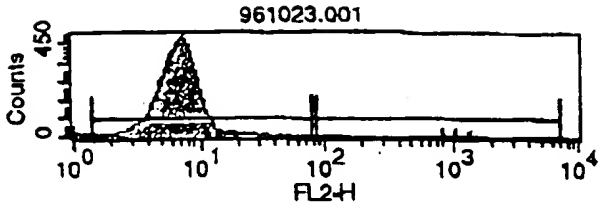
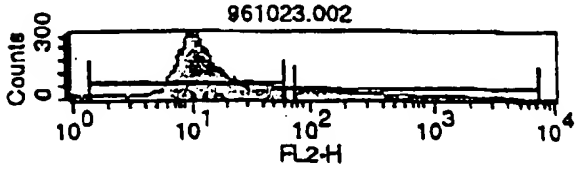
control
9-cis RA
PGJ2+LG268
PGJ2 + LG268+ 109

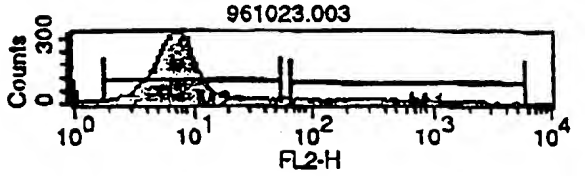
FIG. 2



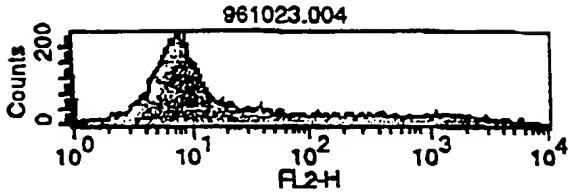
Marker	Mean	Geo Mean	CV	Median
All	7.01	6.53	38.21	6.73
M1	***	***	***	***
M2	7.05	6.61	37.49	6.73



Marker	Mean	Geo Mean	CV	Median
All	73.39	20.52	315.72	12.86
M1	384.14	235.15	125.20	191.10
M2	15.40	12.76	70.60	11.55



Marker	Mean	Geo Mean	CV	Median
All	53.41	10.44	386.57	7.50
M1	454.48	289.85	109.71	264.16
M2	8.84	7.41	79.63	7.23



Marker	Mean	Geo Mean	CV	Median
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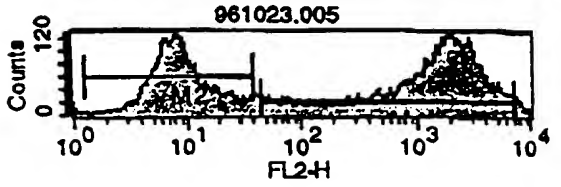

Marker	Mean	Geo Mean	CV	Median
All	1124.70	164.25	134.73	352.27
M1	1809.78	1124.20	80.10	1596.34
M2	10.76	8.87	69.26	8.35

FIG. 3

SEQUENCE LISTING

<110> Evans, Ronald M.
Tontonoz, Peter
Nagy, Laszlo

<120> USE OF RAR ANTAGONISTS AS MODULATORS OF HORMONE MEDIATED PROCESSES

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            20                  25                  30
Xaa Xaa Xaa Xaa Xaa Xaa Cys Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Cys
        35                  40                  45
Xaa Xaa Xaa Lys Xaa Xaa Arg Xaa Xaa Cys Xaa Xaa Cys Arg Xaa Xaa
    50                  55                  60
Lys Cys Xaa Xaa Xaa Gly Met
65                  70
```

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US00/18543

A. CLASSIFICATION OF SUBJECT MATTER

IPC(7) :A61K 7/06, 31/20, 31/23, 31/335, 7/48
US CL :514/569, 568, 532, 63, 544, 682, 549
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

U.S. : 514/569, 568, 532, 63, 544, 682, 549

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

Please See Extra Sheet.

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X - Y	US 5,798,354 A (BERNARDON et al) 25 August 1998, entire text, especially column 7, lines 63-67.	1 ---- 2-29
X - Y	US 5,786,379 A (BERNARDON et al) 28 July 1998, entire text, especially claims 1-56.	1, 13 --------- 2-12 and 14-29
X - Y	US 5,574,036 A (BERNARDON et al) 12 November 1996, entire text, especially claims and column 7, lines 57-67.	1 ----- 2-29
X,P ---- Y,P	US 5,952,382 A (BERNARDON et al) 14 September 1999, entire text, especially claim 1.	1 --- 2-29

[X] Further documents are listed in the continuation of Box C. [ ] See patent family annex.

*	Special categories of cited documents:	"T"	later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
"A"	document defining the general state of the art which is not considered to be of particular relevance	"X"	document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
"E"	earlier document published on or after the international filing date	"Y"	document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
"L"	document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)	"&"	document member of the same patent family
"O"	document referring to an oral disclosure, use, exhibition or other means		
"P"	document published prior to the international filing date but later than the priority date claimed		

Date of the actual completion of the international search 27 JULY 2000	Date of mailing of the international search report 23 AUG 2000
Name and mailing address of the ISA/US Commissioner of Patents and Trademarks Box PCT Washington, D.C. 20231 Facsimile No. (703) 305-3230	Authorized officer VICKIE KIM Telephone No. 703-308-1235

Form PCT/ISA/210 (second sheet) (July 1998)*

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US00/18543

C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	US 5,696,104 A (DEMARCHEZ et al) 09 December 1997, full text, especially claims.	1-29
Y,P	US 6,004,987 A (DEMARCHEZ et al) 21 December 1999, full text, especially column 6, lines 35-51.	1-29
Y,P	US 5,973,007 A (DEMARCHEZ et al) 26 October 1999, entire text, especially claims.	1-29
Y	US 5,827,500 A (DEMARCHEZ et al) 27 October 1998, full text, especially claims.	1-29
Y	US 5,780,676 A (BOEHM et al) 14 July 1998, full text, especially column 61 and 62.	1-29
Y,P	US 6,008,204 A (KLEIN et al) 28 December 1999, entire text, especially abstract and claims.	1-29
Y,P	US 5,998,654 A (BOEHM et al) 07 December 1999, full text, especially claims.	1-29

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US00/18543

B. FIELDS SEARCHED
Electronic data bases consulted (Name of data base and where practicable terms used):

STN, CAPLUS, MEDLINE, USPATFUL, EUROPATFUL, JAPIO, TOXLINE, SCISEARCH, BIOSIS, EMBASE
search terms: RAR antagonist, PPAR agonist, RXR agonist, RAR agonist, steroid/thyroid receptor agonist, permissive receptor, heterodimer